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Synchytrium solstitiale sp. nov. causing a false rust on *Centaurea solstitialis* in France

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Abstract: A new species of *Synchytrium, S. solstitiale,* infecting leaves of *Centaurea solstitialis* in France, is described and illustrated. *Synchytrium solstitiale* causes development of orange to red galls on the leaves and petioles of living plants. It differs microscopically from all previously described species of the genus mainly in having larger sporangia and zoospores and resting spores that are formed in succession without an evanescent prosoral stage.

Key words: Chytridiales, galls, systematics, yellow starthistle

INTRODUCTION

In the search for biological control agents against the noxious weed *Centaurea solstitialis* L. (yellow starthistle) a new parasitic fungus was discovered in a single locality in southern France. Plants were observed to be infected with a species of *Synchytrium* that caused orange galls to form on the leaves and petioles. The taxon on *C. solstitialis* was compared to descriptions of species in *Synchytrium* and determined to be a new species, which is described and illustrated below.

MATERIALS AND METHODS

Leaves of *C. solstitialis* with typical orange galls were collected in Cazevieille, France, (43°43′20″ N; 03°47′42″ E) and immediately processed for sectioning. Symptomatic tissue was excised from the *C. solstitialis* leaf and immersed under vacuum for 4 h in 1 mL of 3% glutaraldehyde in 0.1 M potassium phosphate buffer, pH 7.2. The fixative was removed and the sample was rinsed two times with 0.1 M potassium phosphate buffer, pH 7.2. The samples were dehydrated in a graded series of acetone (10, 20, 30, 50, 70, 90 and 100%) for 10 min. in each wash. The samples then were washed three times in 100% acetone. The samples were infiltrated with Spurr's resin (Dawes 1994) by maintaining them 1 h at room temperature in a mixture of 30% resin and 70% acetone, 4 h in a mixture of 50% resin and 50% acetone, then overnight in a mixture of 70% resin and 30% acetone. After an additional 8 h in 100% resin, the samples were placed in molds, covered with fresh resin and placed in an oven at 70 C for 16 h.

Thick (5 μm) microtome sections were prepared by using a tungsten blade on a Leica RM 2145 microtome (Leica Microsystems, Nussloch, Germany). Sections were stained with a solution of methylene blue-azure A and counterstained with a solution of basic fuchsin (Humphrey and Pittman 1974). The slides were observed under an Olympus Vanox microscope (Olympus, Tokyo, Japan).

Zoospores were induced to release by placing segments of freshly collected infected leaves in a glass well containing sterile distilled water plus 100 mg/L streptomycin. The well was placed in a glass Petri plate with moist filter paper in an incubator at 15 C with artificial light. After 6 h, zoospores were observed and measured.

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TAXONOMY

Synchytrium solstitiale Widmer sp. nov. FIGS. 1–5

Gallae in statu aggregato dihomeogallicae, in superficiebus ambabus foliorum petiolorumque formantes, secus venas abundantes, singulariter vel in gallas multiplices coalescentes, aurantiacae vel atro-rubrae. Prosorus non visus. Sori solitares, sphaerici, cellulam hospitis penitus implentes, 96–152 μm diametro (x = 124 μm, n = 29). Sporangia aurantiaca, figura irregulari, pro parte maxima polyhedrica, (22–72) × (18–56) μm diametro (x = 54 × 37 μm, n = 27), 5–25 per sorum. Sporae quiescentes sphaericae, laeves, olivaceo- vel atro-brunneae, 34–48 μm diametro (x = 42 μm, n = 35). Zoosporae sphaericae, typice globulis aureis duobus, interdum unico vel tribus, 4–8 μm diametro (x = 6 μm, n = 40), flagello unico posteriore 8–16 μm longo (x = 14 μm, n = 20).

Galls compositely dihomeogallic, forming on both sides of the leaves and petioles, abundant along veins, occurring singly or coalescing to form compound galls, orange to dark red. Prosorus not observed. Sori solitary, spherical, filling host cell completely, 96–152 μm diam (x = 124 μm, n = 29). Sporangia orange, irregular in shape, predominantly polyhedral, (22–72) × (18–56) μm diam (x = 54 × 37 μm, n = 27), 5–25 per sorus. Resting spores spherical, smooth, olive brown to dark brown, 34–48 μm diam, (x = 42 μm, n = 35). Zoospores spherical, typically with two gold globules, sometimes one or three globules, 4–8 μm diam (x = 6 μm, n = 40) with single posterier flagellum 8–16 μm long (x = 14 μm, n = 20).

FIG. 1. *Centaurea solstitialis* plant showing typical symptoms of orange galls on a rosette leaf.

Specimen examined. FRANCE. HÉRAULT: Cazevieille-Pic St. Loup, 43°43′20″ N; 03°47′42″ E, on leaves and petioles of *Centaurea solstitialis,* 3 Nov 2000, *T. Widmer,* (HOLOTYPE: BPI 842228).

Etymology. From the Latin *solstitiale,* referred to the plant species from which the species was collected.

Known distribution. France.

Habitat. Parasitic on leaves and petioles of *Centaurea solstitialis*.

Galls protruding from the surface of the leaves and petioles of *C. solstitialis* were mostly singular but also coalesced together giving the appearance of rust pustules, hence the name of the disease ''false rust''. The average gall size was 194 μm diam (n = 20). Some sori also were observed in the trichomes. Heavily infected leaves often were distorted and curled. Normally one or two layers of host cells surrounded the gall.

The genus *Synchytrium* (Chytridiales, Synchytriaceae) is characterized by a holocarpic thallus exogenous to the zoospore cyst, which at maturity develops into a sorus, prosorus or resting spore (Barr 1980). The new species clearly belongs in *Synchytrium* because it lacks true mycelium and forms sporangia and resting spores that release zoospores with a single posterier flagellum. The majority of the species in *Synchytrium* are assigned to two main groups on

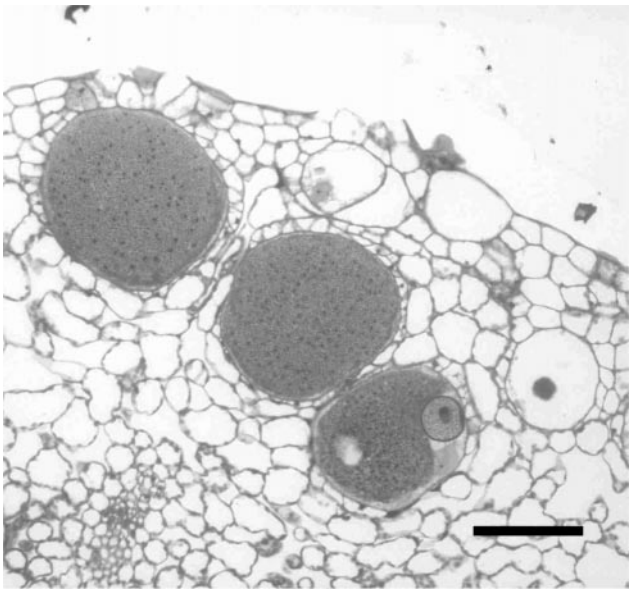

FIG. 2. *Synchytrium solstitiale* sori and resting spore (lower right) embedded in tissue. Bar = 100 μm.

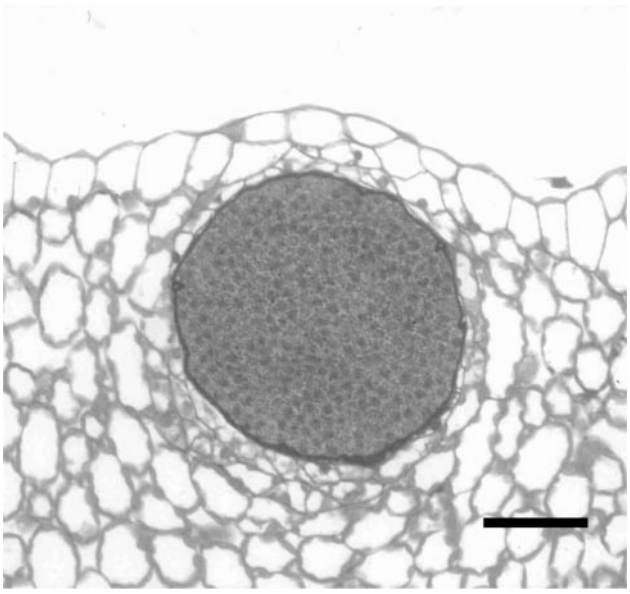

FIG. 3. Single sorus of *Synchytrium solstitiale* embedded in tissue. Bar = 50 μm.

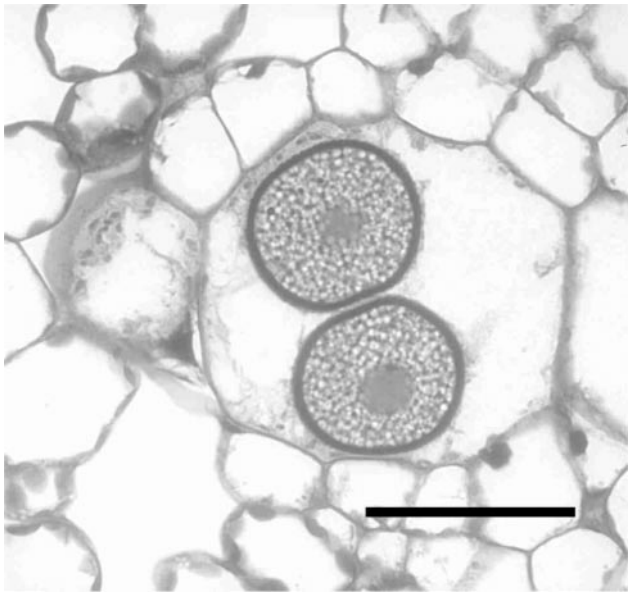

FIG. 4. Two resting spores of *Synchytrium solstitiale* within the same host cell. Bar = 50 μm.

the basis of whether they are long-cycled, forming both sporangial sori and resting spores, or short-cycled, developing only sporangial sori or resting spores. Because *S. solstitiale* is long-cycled with the initial thallus transforming directly into a sorus and has digallic galls that are compositely dihomeogallic, this species is placed in the subgenus *Synchytrium* (*Eusynchytrium*). Karling (1964) reported two species of *Synchytrium, S. aureum* and *S. macrosporum,* on *Centaurea* spp. but not on *C. solstitialis*. Unlike these two species, *Synchytrium solstitiale* is compositely dihomeogallic and has only 5–25 sporangia per sorus. *Synchytrium aureum* and *S. macrosporum* are compositely monogallic and have more than 100 sporangia per sorus. In addition, the resting spores of *S. solstitiale* are 32–48 μm diam while those of *S. aureum*

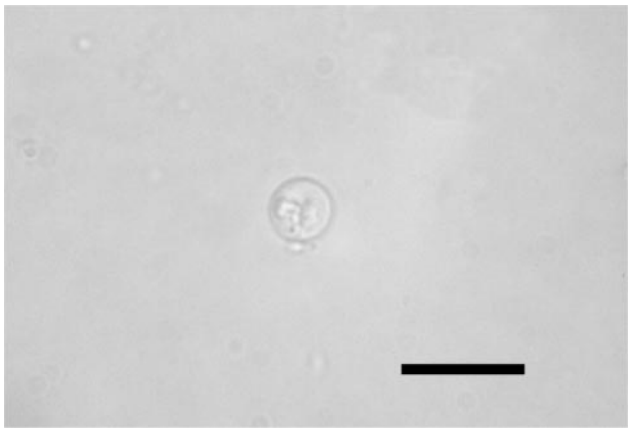

FIG. 5. Single zoospore of *Synchytrium solstitiale*. Bar = 10 μm.

TABLE I. Comparison of *Synchytrium solstitiale* with other *Synchytrium* spp. (Karling, 1964)

Species	Subgenus	Gall type	Sporangia (μm)	Resting spores (μm)	Zoospore (μm)
Synchytrium solstitiale	Synchytrium (Eusynchytrium)	compositely dihomeogallic	57 × 37	32–48	4–8
Synchytrium taraxaci	Synchytrium (Eusynchytrium)	compositely dihomeogallic	45–80	50–80	3.5–4
Synchytrium aureum	Pycnochytrium (Chrysochytrium)	compositely monogallic	17–35	50–200	3–3.2 × 4–4.8
Synchytrium macrosporum	Pycnochytrium (Chrysochytrium)	compositely monogallic	8–35	80–270	3–4 × 4–5
Synchytrium globusum	Leucochytrium	compositely monogallic	14–20	60–170	3–4
Synchytrium endobioticum	Mesochytrium	compositely dihomeogallic	62–78 × 25–38	40–70	1.5–2.2

and *S. macrosporum* are much larger, 50–200 μm and 80–270 μm, respectively. Other species of *Synchytrium* occur on a wide range of hosts in the family Asteraceae. However, all of those species, except *S. taraxaci,* differ in various characteristics such as being compositely monogallic or forming a prosorus, which place them in different subgenera (TABLE I). *Synchytrium taraxaci,* the only other species fully known and placed with certainty in the subgenus *Synchytrium* (*Eusynchytrium*), has larger resting spores (50–80 μm diam) and smaller zoospores (3.5–4 μm diam) than *S. solstitiale*. *Synchytrium endobioticum,* the causal agent of potato wart, differs from the newly reported species in that the sorus of *S. endobioticum* forms above a prosorus, the sorus generally is smaller, and the zoospores are smaller than those structures in *S. solstitiale*. A prosorus was not observed in *S. solstitiale*.

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LITERATURE CITED

Barr, DJS. 1980. An outline for the reclassification of the Chytridiales, and for a new order, the Spizellomycetales. Can J Bot 58:2380–2394.

Dawes, CJ. 1994. Introduction to biological electron microscopy: theory and techniques. Burlington, Vermont: Ladd Research Industries, Inc. 315 p.

Humphrey, CD. and Pittman, FE. 1974. A simple methylene blue-azure II-basic fuchsin stain for epoxy-embedded tissue sections. Stain Technol 49:9–14.

Karling, JS. 1964. *Synchytrium*. New York: Academic Press. 470 p.